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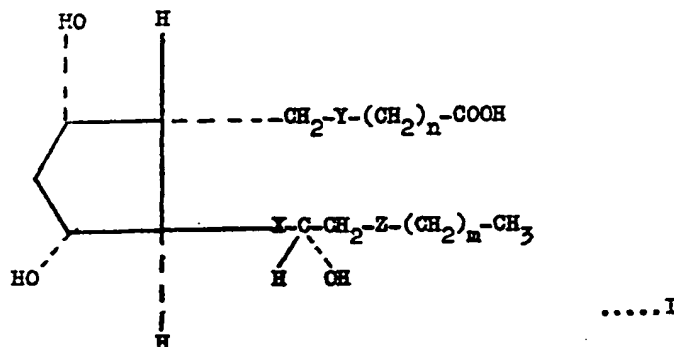
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CONTROL OF FEMALE MAMMALS

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PGF_{1α} is U.S. Patent No. 3,069,322. Bergstrom, Carlson and Weeks, Pharmacological Reviews, Vol. 20, N. 1, 1892 (1968) review "The Prostaglandins".

It has now been found according to the present invention that a method of ensuring the regularity of menses of an ovulating female mammal comprises administering systemically to the mammal a compound of the formula: —

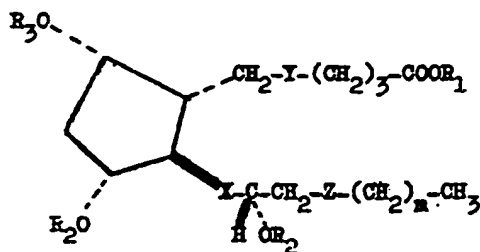


wherein X is CH₂CH₂ or trans CH=CH and both Y and Z are CH₂CH₂; X is trans CH=CH, Y is cis CH=CH and Z is CH₂CH₂ or cis CH=CH; m is 0, 1 or 2 and n is 2, 3, 4 or 5 or an acylate thereof wherein the or each acyl radical is that of a hydrocarbon carboxylic acid having 1 to 8 carbon atoms, or a pharmaceutically acceptable salt or carboxylate ester derived from a hydroxy compound having 1 to 8 carbon atoms inclusive of such a compound, on one or more occasions during a period starting substantially at ovulation and ending at the anticipated menses.

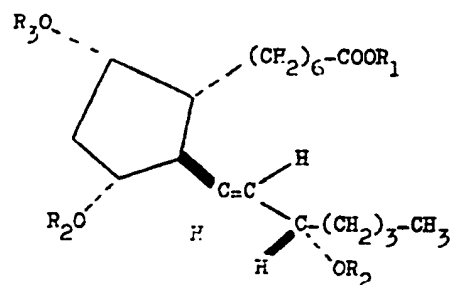
Pharmaceutically acceptable salts for example, those of alkali metals and alkaline earth bases, such as the sodium, potassium, calcium and magnesium salts; those of ammonia or a basic amine such as mono-, di-, and triethylamines, benzylamine, heterocyclic amines such as piperidine and morpholine, and amines containing water-solubilizing or hydrophilic groups such as triethanolamine and phenylmonoethanolamine are disclosed in U.S. Patent No. 3,296,091. Carboxylate esters such as methyl, ethyl, and cyclohexyl having no more than 8 carbon atoms are formed by the usual methods, e.g. reaction with dimethylmethane or similar diazohydrocarbons as in U.S. Patent No. 3,296,091. Acylates of lower alkanolic acids of 1 to 8 carbon atoms inclusive are prepared in the usual manner by reaction of the respective prostaglandin acids with the appropriate acid anhydride or acid halide, e.g., those of acetic, propionic, butyric, isobutyric, valeric, caproic, and caprylic acids, as in Great Britain Patent Specification No. 1,040,544.

The regularity of menses can be ensured by the methods of invention even if the female mammal has been exposed to a male. Thus pregnancy of a female mammal that has been exposed to a male at or subsequent to ovulation is prevented by administering systemically to the mammal on one or more occasions subsequent to exposure but prior to the anticipated menses a compound of Formula I.

Compounds which are especially useful for the above purposes are those having the general formulae: —



and



.....III

wherein R_1 is hydrogen, alkyl of 1 to 8 carbon atoms inclusive, or a pharmacologically acceptable cation, R_2 and R_3 are hydrogen or alkanoyl of 1 to 8 carbon atoms inclusive with the proviso that when R_3 is alkanoyl, R_2 is also alkanoyl, m is zero or 2, and X , Y and Z are $-\text{CH}_2\text{CH}_2-$, or X is $\text{trans}-\text{CH}=\text{CH}-$, Y is $\text{cis}-\text{CH}=\text{CH}-$, and Z is $-\text{CH}_2\text{CH}_2-$ or $\text{cis}-\text{CH}=\text{CH}-$.

When R_2 and R_3 in a compound of Formula II or III are both alkanoyl they can be the same or different.

These compounds of Formulae II and III and the preparation thereof are described and claimed in our copending Application No. 8645/72 (Serial No. 1285372).

Examples of alkyl of 1 to 8 carbon atoms are methyl, ethyl, propyl, butyl, pentyl, hexyl, heptyl, octyl, and isomeric forms thereof.

Examples of alkanoyl of 1 to 8 carbon atoms, inclusive are formyl, acetyl, propionyl, butyryl, valeryl, hexanoyl, heptanoyl, octanoyl, and isomeric forms thereof.

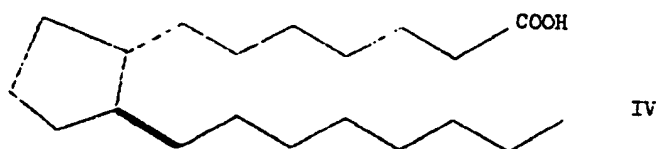
Pharmacologically acceptable cations within the scope of R_1 in formulas II and III are quaternary ammonium ions or the cationic form of a metal, ammonia, or an amine.

Especially preferred metal cations are those derived from the alkali metals, e.g., lithium, sodium, and potassium, and from the alkaline earth metals, e.g., magnesium and calcium, although cationic forms of other metals, e.g., aluminium, zinc, and iron, are also within the scope of R_1 .

Pharmacologically acceptable amine cations within the scope of R_1 in formulas II and III are those derived from primary, secondary, or tertiary amines. Examples of suitable amines are methylamine, dimethylamine, trimethylamine, ethylamine, dibutylamine, triisopropylamine, N-methylhexylamine, decylamine, dodecylamine, allylamine, crotylamine, cyclopentylamine, dicyclohexylamine, benzylamine, dibenzylamine, α -phenylethylamine, β -phenylethylamine, ethylenediamine, diethylenetriamine, and like aliphatic, cycloaliphatic, and araliphatic amines containing up to and including about 18 carbon atoms, as well as heterocyclic amines, e.g., piperidine, morpholine, pyrrolidine, piperazine, and lower-alkyl derivatives thereof, e.g., 1-methylpiperidine, 4-ethylmorpholine, 1-isopropylpyrrolidine, 2-methylpyrrolidine, 1,4-dimethylpiperazine, and 2-methylpiperidine, as well as amines containing water-solubilizing or hydrophilic groups, e.g., mono-, di-, and triethanolamine, ethyldiethanolamine, N-butylethanolamine, 2-amino-1-butanol, 2-amino-2-ethyl-1,3-propanediol, 2-amino-2-methyl-1-propanol, tris(hydroxymethyl)aminomethane, N-phenylethanolamine, N-(*p*-tert-amylphenyl)diethanolamine, galactamine, N-methylglucamine, N-methylglucosamine, ephedrine, phenylephrine, epinephrine, and procaine.

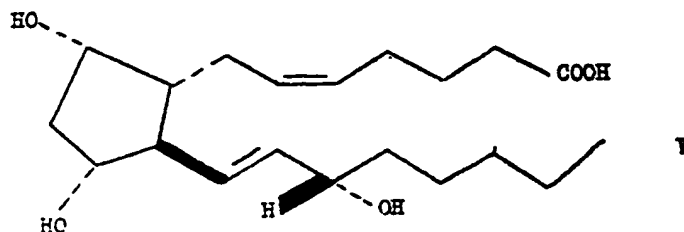
Examples of suitable pharmacologically acceptable quaternary ammonium cations within the scope of R_1 in formulas II and III are tetramethylammonium, tetraethylammonium, benzyltrimethylammonium, and phenyltriethylammonium.

The compounds of formulas II and III are somewhat similar to certain of the natural prostaglandins. The latter are considered to be derivatives of prostanoic acid which has the following structure:



IV

The naturally-occurring prostanoid acid derivative, prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), has the following structure:



The compound of formula II wherein R_1 , R_2 , and R_3 are hydrogen, and X is trans-CH=CH- , Y is cis-CH=CH- , and Z is $\text{-CH}_2\text{CH}_2\text{-}$, has the same structure as $PGF_{2\alpha}$, except that this formula II compound has one less carbon atom in the hydroxy-containing side chain (ω -nor) when m is zero, and one more carbon atom in the same chain (ω -homo) when m is 2. The other compounds encompassed by formula II are similarly related to the known prostanoid acid derivatives dihydro- $PGF_{1\alpha}$ and $PGF_{3\alpha}$. The compound of formula III wherein R_1 , R_2 , and R_3 are hydrogen has one less carbon (ω -nor) than the known $PGF_{2\alpha}$.

These ω -nor and ω -homo $PGF_{2\alpha}$ compounds of formulas II and III are extremely potent in causing various biological responses of the general type caused by the corresponding natural $PGF_{2\alpha}$ compounds. Regarding the biological responses caused by the natural $PGF_{2\alpha}$ compounds, see, for example, Bergstrom et al., *Pharmacol. Rev.* 20, 1 (1968), and references cited therein.

Thus these formula II and III compounds are useful for ensuring the irregularity of menses and in place of oxytocin to induce labor in pregnant animals, including man, cows, sheep, and pigs, at or near term, or in pregnant animals with intrauterine death of the fetus from about 20 weeks to term. For this purpose, the compound is preferably infused intravenously at a dose 0.01 to 50 $\mu\text{g.}$ per kg. of body weight per minute until or near the termination of the second stage of labor, i.e., expulsion of the fetus. These compounds are especially useful when the female is one or more weeks post-mature and natural labor has not started, or 12 to 60 hours after the membranes have ruptured and natural labor has not started.

Thus a method according to the present invention of inducing labour in a pregnant female mammal comprises administering systemically to the mammal a compound of Formula II or III.

The methods of the invention are preferably conducted by administering the active ingredient systemically to the female mammals, in the form of pharmaceutical compositions. Pharmaceutical compositions comprising as the active ingredient a compound of Formula II or III are described and claimed in our copending Application No. 8645/72 (Serial No. 1285372).

It is preferred to administer the compounds of Formula I in the form of compositions in dosage unit forms for ease and economy of administration and uniformity of dosage. Dosage unit form as used in the specification and claims herein refers to physically discrete units suitable as unitary dosages for animal and human subjects, each unit containing a predetermined quantity of active material calculated to produce the desired biological effect in association with the required pharmaceutical means. The specifications for the dosage unit forms of this invention are dictated by and directly dependent on (a) the unique characteristics of the active material and the particular biological effect to be achieved and (b) the limitations inherent in the art of compounding such an active material for administration to animal and human subjects as disclosed in detail in this specification.

For example effectiveness in ensuring regularity of menses is dependent on providing in the female an effective amount of the active ingredient during a span of time starting approximately at the time of ovulation and ending approximately at the time of menses or just prior to menses. Within this span wherein the preparations and methods are operable, variations in time and frequency of administration are possible provided an effective amount of the essential active ingredient is made available. This span correlates with development of the *corpus luteum* upon which, according to some experimental data, a luteolytic effect is exerted by the present preparations and methods. Various methods of administration are possible. Illustratively, daily intra-

venous infusion of a sterile aqueous pharmaceutical preparation containing the active ingredient starting on or about the sixteenth day of the cycle and ending on or about the last day or two of the cycle is an effective mode of administration. This mode may be varied to allow for infusion of larger amounts on each of two or three days. Infusion administration on the second or third day prior to expected menses can be used. Another method is the injection of a sterile pharmaceutical unit dosage preparation in an aqueous or oily vehicle form injected over a schedule of about one injection on each of two or three days. Preferably the sterile aqueous and the sterile oil preparation contain up to about 500 mg per millilitre of said active ingredient. Also in another preferred feature when the active ingredient is administered in the form of a sterile aqueous solution the latter can contain up to about 50 mg per millilitre of said active ingredient. A further method is the injection of a sterile aqueous suspension of a carboxylate ester as heretofore described or an acylate as heretofore described. In this method one injection on or about the sixteenth or seventeenth day of the cycle is effective to bring about menses in the ovulating female at the usual time although sexual exposure occurs at the time of ovulation. The sterile aqueous suspension preferably also contains up to about 500 mg per millilitre of said carboxylate ester or acylate. Another method is by use of an intravaginal composition; illustratively, an intravaginal suppository administered once every three days starting on or about the seventeenth day of the cycle until the ensuing menses appears. The intravaginal composition preferably contains up to about 500 mg of the active ingredient.

Yet another method is sublingual or buccal administration of a suitable pharmaceutical preparation whereby the principal active ingredient is directly available to the blood supply and thereby exerts its beneficial effect. One such pharmaceutical preparation held under the tongue until dissolved once or twice daily starting on or about the seventeenth day of the cycle is effective to maintain the required amount of the active prostaglandin type ingredient to prevent pregnancy during the particular cycle although ovulation and exposure to the male have occurred. Other injectables are, for example, combinations of a water soluble salt and an acylate or carboxylate ester to provide both immediate and prolonged action. A dry preparation for reconstitution as desired with an appropriate liquid, e.g. sterile saline is also used in another method. The compound is administered sublingually or buccally preferably in dosage unit form containing up to 200 mg of the active ingredient.

The aforesaid prostaglandins are administered in dosage unit forms of pharmaceutical preparations supplying to the treated female mammal an effective amount of the essential active ingredient for control of the reproductive cycle, i.e. by ensuring a nonpregnant cycle in the female notwithstanding ovulation and contact with a fertile male as by natural coitus during the aforesaid span extending from on or about the time of ovulation to just prior to expected menses. Additionally, the ovulating female obtains regularity of the reproductive cycle by utilizing the preparations and methods of this invention, apparently due to aiding the natural cycle regression of the *corpus luteum*. The preparation can be in the form of a fine powder of 25 microns or less, preferably prepared by air micronization, such powder being used as a vaginal insufflation. The powder can be suitably compounded with a compatible extender, e.g., lactose. Other pharmaceutical preparations in dosage unit form are compounded of the essential prostaglandin active ingredient and pharmaceutical means which adapt the preparation for systemic administration. The pharmaceutical preparations for administration to the humans and animals include those for injectable, sublingual, or buccal and vaginal administration. Those for injectable administration are, for example, sterile aqueous solutions, sterile aqueous suspensions, sterile oily solutions or suspensions, sterile powders for subsequent incorporation into an injectable form by addition of the required sterile vehicle. The solutions or suspensions are compounded with the required pharmaceutical means such as preservatives, suspending and dispersing agents, and isotonic agents, for example, methyl and propyl parabens, sodium chloride, polyethylene glycols, especially polyethylene glycol 4000, sodium carboxymethylcellulose, sodium alginate or polyvinyl pyrrolidone, poly sorbate 80, condensation products of ethylene oxide with fatty acids, for example polyoxyethylene stearate; or with fatty alcohols, for example heptadecaethyleneoxyctanol, or with partial esters, for example polyoxyethylene sorbitol mono-oleate or hexitans derived from sorbitol, for example polyoxyethylene sorbitan mono-oleate. Preservative such as methyl and propyl p-

hydroxy benzoates are incorporated into such suspensions or dispersions. Suspensions in oily media can be prepared by dispersing the active ingredient in an acceptable oily means, for example a vegetable oil such as sesame oil, peanut oil and cottonseed oil. These may contain means to delay adsorption, for example aluminum monostearate. All dosage unit forms for injectable administration must be sterile as is known and practiced in the art.

Preparations for vaginal application include the essential active ingredient reduced in particle size to a powder suitable for insufflation or suitably mixed with inert excipient means such as lactose. Such preparations also include suppositories and other formed structures such as ring devices for intravaginal use containing the essential active ingredient, for example a silicone polymer device in the form of a toroid which will release the essential active ingredient during a predetermined period of time. The amount of the essential active ingredient provided by the various dosage forms is sufficient to supply a dosage of from 0.01 mg. to 20 mg. per kilo of the treated host, depending on the desired promptness, duration and magnitude of the end result. The amount of the prostaglandin compound used in the several methods of the invention, whether for oral, injectable, or intravaginal administration can be expressed in percentage by weight or in specific amounts. These percentages or specific amounts will vary in view of the different onset and duration of the biological effects that attend each dosage unit form. For example, a sterile aqueous suspension designed for prolonged action after one injection can contain as much as 50% by weight whereas a sterile aqueous solution as diluted with sterile saline for infusion can contain as little as 0.00005% (0.5 mg. in a 1000 ml. infusion equivalent to 0.01 mg./kilo for a 50 kilogram woman). A sterile aqueous solution for direct intravenous administration, without dilution as with physiological saline can contain for example, 5% or more. Dosage unit forms such as the sublingual and intravaginal types can contain as much as 200 mg. and 500 mg. respectively. Other suitable compositions include, for example, oily preparations, dry preparations for suspension and solution, which are designed to provide the dosages of from 0.01 mg. to 20 mg. per kilo of body weight.

Although the exact mechanism of action of the essential active ingredient of the prostaglandin type in controlling the reproductive cycle is not certain, the action manifests itself in several ways, for example, by regulating menses or heat so that the length of the cycle conforms to a predetermined span; by preventing reproduction despite ovulation and natural exposure to sperm; and by a luteolytic phenomenon involving regression of *corpora lutea*. This phenomenon will terminate anestrus.

The mechanism of action of the prostaglandins in the treated females is a matter for conjecture although experimental data indicate that a luteolytic mechanism and regression of the *corpus luteum* may be involved. A pharmaceutical preparation is made up by dissolving $\text{PGF}_{2\alpha}$ in physiological saline at a concentration of 125 mcg./ml. and adjusting the pH within the range of 5 to 7 with bicarbonate buffer. Cycling normal rats (200 to 300 gm.) are prepared with a right uterine cornea indwelling catheter, Weeks and Davis, J. Appl. Physiol. 19, 540 (1964). At the third normal proestrus pseudopregnancy is induced by vaginal stimulation with an electric probe. Vaginal smears are taken to confirm pseudopregnancy. On the morning of day 5 of pseudo-pregnancy the pharmaceutical preparation of $\text{PGF}_{2\alpha}$ is infused at the rate of 2.06 ml./day equivalent to about 1 mg./kg./day of the $\text{PGF}_{2\alpha}$. The infusions of the $\text{PGF}_{2\alpha}$ preparation and a like saline control are continued for 48 hours at which time the animals are sacrificed and the ovaries harvested and placed in 1 ml. of 2.5% NaOH solutions for determination of progesterone and $20\alpha\text{-OH}$ progesterone. The determinations in two experiments are as listed in Table 1.

TABLE 1

The Effect of PGF_{2α} Infusion on the Concentration of Progesterone and 20αOH—Progesterone in the Ovaries of Pseudopregnant Rats

Sample	Treatment	No. Ovaries	Ovarian Location (side)	Total Weight (mgs)	Progesterone (μg/gm tissue)	20αOHP (μg/gm tissue)	P/OHP Ratio
Experiment 1							
1	Saline (2.06 ml./day) infused into right uterine horn	2	Right	70.0	14.8 } 11.7* 8.6 }	1.40 } 1.2 0.90 }	9.75
2		2	Left	67.3			
3	PGF _{2α} (1 mk/kg/day) into right uterine horn	3	Right	129.8	4.1 } 2.8 2.8 } 3.0 } 1.4 }	18.7 } 11.9 4.1 } 13.6 } 11.5 }	0.24
4		2	Right	94.9			
5		3	Left	150.9			
6		2	Left	89.7			
Experiment 2							
1	Saline (2.06 ml/kg/day) into right uterine horn	3	Right	291.4	4.12 } 5.56 7.02 } 4.61 } 6.47 }	4.80 } 4.9 5.00 } 5.60 } 4.10 }	1.13
2		3	Right	203.5			
3		3	Left	182.8			
4		3	Left	280.3			
5	PGF _{2α} (1 mk/kg/day) into right uterine horn	3	Right	164.8	0.87 } 0.67 0.39 } 0.53 } 0.90 }	13.9 } 10.1 7.9 } 9.0 } 9.4 }	0.06
6		3	Right	218.5			
7		3	Left	131.4			
8		3	Left	207.1			

* Average values for each group.

The data show that the effect of the $\text{PGF}_{2\alpha}$ administration is a reduction of progesterone content and an increase in the reduced steroid content, thus indicating a luteolytic action through failure of effective progesterone content.

Reference is directed under Section 9 to British Patent Specifications Nos. 1,198,071 and 851,827.

The following are some Examples of methods of the invention and compositions for use in these.

EXAMPLE 1

Intravenous infusion of pharmaceutical preparation

$\text{PGF}_{2\alpha}$ is made up in sterile saline solution at a concentration of 0.5 mg./ml. and used for administration by infusion in female rats. Spartan Sprague-Dawley rats are used. Males are experienced breeders and females (225—275 gm. body wt.) have typical vaginal estrus cycles. Indwelling right heart cannulas are inserted during proestrus. After cannulation, daily vaginal smears are again taken to insure maintenance of normal cyclicity. Initially infusion of $\text{PGF}_{2\alpha}$ (3.2 mg./kg./day in saline) commences at 4.00 p.m. the afternoon before mating and continues for six additional days. The starting time of the infusion is later modified to commence on the morning following mating because of an adverse affect on mating behavior of the environment associated with the infusion equipment. The day of finding sperm in the vagina is considered Day 1 and the males are removed from the females at this time.

On Day 8, an exploratory laparotomy is performed under ether anesthesia via a abdominal midline incision. Uteri are checked for number, size, and distribution of implantation sites taking care to minimize any handling of the reproductive tract. Incisions are closed with surgical silk, and animals returned to their original cages. On Day 18 females are placed in casting boxes. At parturition or on Day 23 animals are sacrificed and the number and condition of the young are determined.

Results:

Six of eight rats infused with saline only conceive. Those animals average 11.7 implantation sites at Day 8, and 7.8 develop fetuses.

Three of 11 $\text{PGF}_{2\alpha}$ treated rats conceive. Implantation sites of one of these three are barely detectable at Day 8, no indication of pregnancy is evident at autopsy on Day 23. The remaining two have implants of normal size, on the low side of the average number and, in one rat, are predominantly in the anterior half of the uterine cornu. Five fetuses at term appear normal by gross inspection.

EXAMPLE 2

Subcutaneous administration of pharmaceutical preparation

Prostaglandin ($\text{PGF}_{2\alpha}$) is made up in sterile saline solution at a concentration of 0.8 mg./ml. and used for subcutaneous administration in female rats. Spartan Sprague-Dawley rats are used. Males are experienced breeders and females (225—275 gm.) have typical estrous cycles. Males are placed with females during proestrus and allowed to remain overnight. The following morning females are examined for vaginal plugs and the presence of sperm. Animals with vaginal sperm are started on test, the day of finding sperm being considered Day 1.

A. 3.2 mg./kg. of $\text{PGF}_{2\alpha}$ is injected subcutaneously daily in two evenly divided dosages. Animals are sacrificed on Day 8 at which time the number and size of implants are recorded. The results are in Table 2.

B. Females are injected subcutaneously, b.i.d., (twice daily) on days 4, 5 and 6 with either 0.1 mg., 0.2 mg., 0.4 mg., or 0.8 mg. of $\text{PGF}_{2\alpha}$ per day. The rats are sacrificed on Day 15. At the time of sacrifice the number, size and distribution of implants are recorded. The results are in Table 3.

TABLE 2
Effect of PGF_{2α}, Injected Subcutaneously

Treatment	Dose b.i.d.	Days Injected	Total Dose (mg)—No. of rats	No. of rats with implantation sites	Average No. of Implants
Physiological Saline	0.5 cc	Day 1—7	0.0 (5)	4	11.2
PGF _{2α}	0.4 mg/0.5 cc	Day 1—7	5.6 (5)	0	—
	"	Day 3—	4.0 (4)	0	—
	"	Day 5—7	2.4 (4)	1	13
	"	Day 6—7	1.6 (4)	3	14
	"	Day 6	0.8 (2)	2	8
	"	Day 7	0.8 (2)	2	14.5
	"	Day 1—7	5.6 (3)	0	—
PGF _{2α}	"	Day 1—5	4.0 (4)	2	11.5
	"	Day 1—3	2.4 (4)	4	13
	"	Day 2—4	2.4 (4)	3	7.6
	"	Day 3	0.8 (2)	2	8.5
	"	Day 2	0.8 (1)	1	12

TABLE 3

Effect of PGF_{2α} when injected Subcutaneously n Days 4, 5 and 6

Treatment	Dose b.i.d.	Total Dose (mg)—No. of rats	Day 15	
			No. of rats with implantation sites	Av. No. of Implants
Physiological Saline	0.5 cc	0.0 (3)	3	12.7
	0.05 mg	0.3 (3)	2	10.5
	0.1 „	0.6 (3)	2	10.0
	0.2 „	1.2 (3)	2	5.0
	0.4 „	2.4 (3)	0	—

EXAMPLE 3

Subcutaneous administration of pharmaceutical preparation

PGF_{2α} is made up in sterile physiological saline at a concentration of 10 mg./ml. and used for subcutaneous administration in female rabbits.

Mature virgin Dutch rabbits weighing about 1.5 kg. each are used. Each of ten females is mated twice with two different proven males and the day of finding sperm in the vaginas of the female rabbits is Day 1. Thereafter, subcutaneous injections are begun on Day 4 with the mated animals divided into five groups each.

In Group I each rabbit receives two subcutaneous injections per day of the pharmaceutical preparation providing a total daily dosage of 5 mg./kg./day of the PGF_{2α}. In Group II each of the five mated female rabbits receives two like daily injections subcutaneously 0.5 ml. of physiological saline.

The injections are given on each of five days and thereafter on Day 12 the animals are sacrificed and autopsied. In none of the rabbits injected with the PGF_{2α} preparation are implantation sites found at autopsy. In the other group of saline-treated controls four of the rabbits have implantation sites with the number of implants being respectively 8, 5, 8 and 8. The fifth animal in this group shows no evidence of implantation sites.

EXAMPLE 4

Intravaginal administration of suppository

PGF_{2α} is made up in a suppository base containing two parts by weight of polyethylene glycol 6000 and one part by weight of polyethylene glycol 1500. The suppositories are formed into pellets with a volume of approximately 1 ml. The suppositories for administration of the prostaglandin contain 8 mg. each of the PGF_{2α} prostaglandin material.

Ten female rabbits are each mated twice with two different proven males and the day of finding of sperm in the vagina is taken as Day 1. Each rabbit is treated once on days 4, 5, 6, 7, and 8 for a total of five days. Since the individual rabbits weight about 1.6 kg., the dosage of the prostaglandin material in the medicament-treated animals is 5 mg./kg./day.

In none of the five prostaglandin-treated rabbits are implantation sites found upon autopsy on Day 12. In each of the five control rabbits treated with saline there are implantation sites averaging 6, 5, 5, 7 and 9 sites respectively.

EXAMPLE 5

Aqueous solution

Mature female rhesus monkeys (5—6 kg.) are mated naturally at a time and for a duration of the reproductive cycle calculated to maximize the chances of conception. The day of vulvation is determined by following peripheral blood progesterin levels, and ovulation is confirmed by laparotomy. Prostaglandin F_{2α} (PGF_{2α}) is dissolved 25 mg./ml. in ethanol and diluted to 15 mg./ml. with a sterile aqueous methylcellulose

vehicle (0.25%). This prostaglandin preparation is injected subcutaneously b.i.d., 30 mg./day for 5 days. Injection is initiated on Day 7 after the presumed day of ovulation in one animal, female No. 16-M. The other three test animals are injected on Days 11 to 15 post ovulation. Peripheral plasma progesterone levels are followed during the cycle current to the time of injection. Pregnancy is diagnosed by rectal palpation to determine uterine enlargement. All test animals are observed for systemic signs of drug toxicity during the course of the experiment.

Peripheral blood progesterone levels are not completely depressed by initiating prostaglandin injection on Day 7. However, progesterone levels fall precipitously almost to non-detectable levels in three test animals following the initiation of drug injection on Day 11. This drop in progesterone level is followed by onset of menses on the 2nd, 3rd and 4th day of injection. One of the four test animals, No. 2-M is diagnosed pregnant 40 days after mating. Previous control fertility and a confirmed pregnancy in a concurrent control animal indicate planned mating under the conditions of this experiment results in a 75—80% fertility rate.

No systemic signs of drug toxicity are noted in any of the animals in the present experiment. Slight tissue necrosis at some of the injection sites and a general tightening of the skin in the local area of injection are noted.

EXAMPLE 6

Sterile Aqueous Suspension

A sterile vehicle is prepared to contain in each milliliter 30 mg. of polyethylene glycol 400 U.S.P. and 2.9 mg. of preservative. Sterilization is accomplished by filtration thru a sterile clarifying pad.

2.2 liters of suspension is prepared to contain 400 mg. per ml. of the acetate of PGF_{1α}.

Acetate of PGF _{1α} , Sterile, micronized	Each ml.	Total
Sterile Vehicle	400 mg.	898 Gm. 1496 Gm.

Add the sterile acetate to about 95% of the required vehicle until a smooth suspension is obtained. Add the balance of the sterile vehicle and mix well. Pass the whole thru a sterile mill and collect in a sterile container.

Intramuscular injection of 1 ml. to the ovulating human 1 day after coitus during the fertile period is followed by menses at the usual time.

The acetate is replaced by the propionate or aforesaid similar acylate or by the methyl, ethyl or similar ester of PGF_{1α} with like results.

EXAMPLE 7

Sterile Aqueous Solution

A sterile aqueous solution for intravenous infusion administration is prepared from the following ingredients to contain 25 mg. per ml. of the sodium salt of PGF_{2α}.

Sodium PGF _{2α}	25 Gm.
Lactose Hydrous	50 Gm.
Sodium Biphosphate anhydrous	1.6 Gm.
Sodium Phosphate Exsiccated	17.5 Gm.
Water for injection q.s. ad	1000 ml.

One milliliter is administered by intravenous infusion to the ovulating human female after intercourse during the fertile period of the cycle. The infusion is given two days before expected onset of menses. It can be repeated on the day before expected menses. Lesser amounts of the active ingredient can be used for infusions given on three or four days or on several days. Thereafter menses occurs at the usual time in the menstrual cycle.

EXAMPLE 8

Intravaginal Suppository

Intravaginal suppositories are prepared to contain in each suppository 250 mg. of prostaglandin PGF_{2α}. One thousand suppositories are prepared by moulding a mixture of the following ingredients:

PGF _{2α} , micronized	250 Gm.
Polyethylene glycol 6000	650 Gm.
Lactose	100 Gm.

5 Starting on the second day post-ovulation one suppository is used intravaginally each day in the ovulating human female with the result that menses occurs on the 28th day of a normal 28-day menstrual cycle. 5

EXAMPLE 9 Intravaginal Device

10 An intravaginal device in the form of a toroid is prepared to contain 700 mg. of dihydro PGF_{1α} dispersed in the toroid. The prostaglandin-type active ingredient is dispersed throughout a vulcanizable polysiloxane polymer which is then moulded in a ring structure to provide a toroid for placement in the vaginal tract. The toroid ring structure is inserted into the vagina after ovulation where it releases the active ingredient and exerts its beneficial biological effect with menses following at the expected time in the menstrual cycle. At that time the intravaginal device is removed. Like results are obtained with an annular ring coated with the prostaglandin. 15

EXAMPLE 10 Sublingual Administration

20 One thousand tablets are prepared from the following ingredients, each containing 50 mg. of active ingredient. 20

PGF _{2α} , micronized	50 Gm.
Polyethylene glycol 4000, powdered	150 Gm.
Polyethylene glycol 6000, powdered	75 Gm.

25 The materials are mixed well and compressed into sublingual-type tablets of the proper weight. At the time of ovulation the human female uses one under the tongue and one daily thereafter to ensure that menses will occur at the end of the normal menstrual cycle in the particular female. 25

EXAMPLE 11 Sterile Aqueous Solution

30 A sterile aqueous solution containing in each milliliter 50 mg. of PGF_{2α} is prepared from the following ingredients: 30

PGF _{2α}	50 Gm.
Ethanol	300 ml.
Water for injection q.s. ad	1000 ml.

35 The PGF_{2α} is dissolved in the ethanol and then carefully diluted with the sterile water for injection. Thereafter the whole is sterilized by sterile filtration. One milliliter injected intravenously into an ovulating bitch on the 10th and 15th days after sexual contact with a known fertile stud is beneficial in insuring that the usual heat period will take place in the bitch indicating that pregnancy is prevented. 35

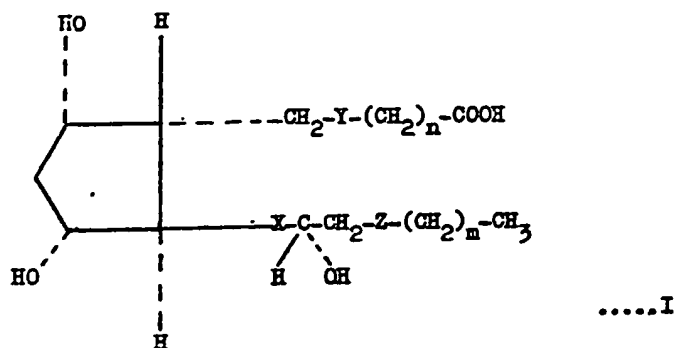
40 Other embodiments in the various dosage unit forms are prepared with the additional compounds represented by the formula heretofore described and used with like beneficial results in the control of the reproductive cycle. 40

45 The claims that are set out below are solely claims to methods of treating female mammals and we specifically state that no claim is made to the mammals treated by these methods. 45

We make no claim herein to any method of treating a human ovulating female in a manner contrary to the offences against the Person Act 1861, as amended and clarified by the Abortion Act 1967. Subject to the aforesaid disclaimers:—

WHAT WE CLAIM IS:—

50 1. A method of ensuring the regularity of menses of an ovulating female mammal comprising administering systemically to the mammal a compound of the formula:— 50



wherein X is CH_2CH_2 or trans $\text{CH}=\text{CH}$ and both Y and Z are CH_2CH_2 ; X is trans $\text{CH}=\text{CH}$, Y is cis $\text{CH}=\text{CH}$ and Z is CH_2CH_2 or cis $\text{CH}=\text{CH}$; m is 0, 1 or 2 and n is 2, 3, 4 or 5 or an acylate thereof wherein the or each acyl radical is that of a hydrocarbon carboxylic acid having 1 to 8 carbon atoms, or a pharmaceutically acceptable salt or carboxylate ester derived from a hydroxy compound having 1 to 8 carbon atoms inclusive of such a compound, on one or more occasions during a period starting substantially at ovulation and ending at the anticipated menses.

2. A method in which pregnancy of a female mammal that has been exposed to a male at or subsequent to ovulation is prevented by administering systemically to the mammal on one or more occasions subsequent to exposure but prior to the anticipated menses a compound as defined in claim 1.

3. A method according to claim 1 in which the compound is administered by intravenous infusion to the mammal each day from substantially the sixteenth day of the menstrual cycle.

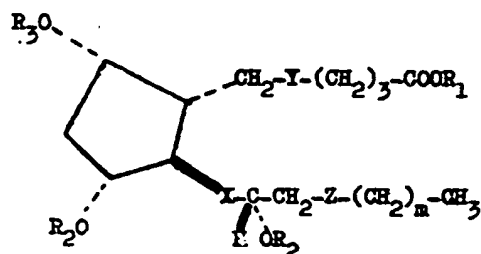
4. A method according to claim 1 in which the compound is administered in the form of an intravaginal composition once every three days from substantially the seventeenth day of the menstrual cycle.

5. A method according to claim 1 in which the compound is injected in the form of a sterile aqueous or oily suspension on two or three days of the period.

6. A method according to claim 1 in which the compound is administered by infusion two or three days before the anticipated menses.

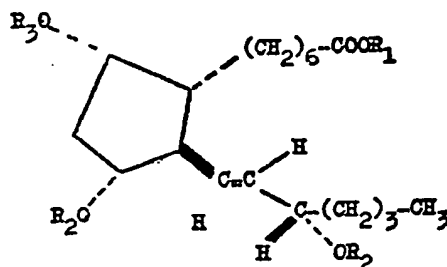
7. A method according to claim 1 or 2 in which a sterile aqueous dispersion of an acylate or carboxylate ester as defined in claim 1 is administered by injection substantially on the sixteenth or seventeenth day of the menstrual cycle.

8. A method according to any preceding claim in which the compound is a compound of the formula:—



wherein R_1 is hydrogen, alkyl of 1 to 8 carbon atoms, inclusive, or a pharmacologically acceptable cation, R_2 and R_3 are hydrogen or alkanoyl of 1 to 8 carbon atoms, inclusive with the proviso that when R_3 is alkanoyl, R_2 is also alkanoyl, m is zero or 2, and X, Y, and Z are $-\text{CH}_2\text{CH}_2-$, or X is trans $-\text{CH}=\text{CH}-$, Y is cis $-\text{CH}=\text{CH}-$, and Z is $-\text{CH}_2\text{CH}_2-$ or cis $-\text{CH}=\text{CH}-$.

9. A method according to any of claims 1 to 7 in which the compound is a compound of the formula:—



wherein R_1 is hydrogen, alkyl of one to 8 carbon atoms, inclusive, or a pharmacologically acceptable cation, and R_2 and R_3 are hydrogen or alkanoyl of one to 8 carbon atoms, inclusive, with the proviso that when R_3 is alkanoyl, R_2 is also alkanoyl.

5 10. A method of inducing labour in a pregnant female mammal comprising administering systemically to the mammal a compound as defined in claim 8 or claim 9.

11. A method according to claim 5 or 7 in which the sterile aqueous preparation or suspension contains up to about 500 mg per millilitre of said compound.

10 12. A method according to claim 5 in which the sterile oil preparation contains up to about 500 mg per millilitre of said compound.

13. A method according to claim 4 in which the intravaginal composition contains up to about 500 mg of the compound.

14. A method according to claim 1 in which the compound is administered sublingually or buccally in dosage unit form containing up to 200 mg of said compound.

15 15. A method according to claim 5 in which the sterile aqueous solution contains up to about 50 mg per millilitre of said compound.

16. A method according to any previous claim in which the compound administered is $\text{PGF}_{2\alpha}$.

20 17. A method according to any preceding claim substantially as herein described with reference to the Examples.

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 DT-01-561. R35.
 Hester K. BE--740615-Q.

A61K (31-07-70)...
 HAIR LOSS PREVENTION AGENT...

NEW

Method of preventing the falling-out of hair characterized by applying a composition containing mineral salts associated with a calcium-releasing agent e.g. parathyroid hormone or dihydroxycholesterol and a capillary-permeability inhibitor.

ADVANTAGE

Application of the compositions to the scalp decreases the permeability of the capillaries unlike usual treatments in which a vasodilator is used. The nutrition of the scalp is improved.

DETAILS

Mineral salts may be those of calcium and/or potassium. Mg salts may also be used; preferred anions are phosphates, PO_4^{3-} or SO_4^{2-} . Compositions may be improved by including a detoxifying agent especially acetylmethionine or EACA. Preferred inhibitors of capillary

RA-G, B4-B25, 5, A1A, 1, A1B, 10-A1,
 B10-E2, B12-...

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permeability or permeabilities of ratio of blood to skin.

EXAMPLE

A hair-dressing composition contained parathyroid hormone 20 units, KCl 50 mg, $MgCl_2$ 38.1 mg, $CaCl_2$ 44.4 mg, D-acetylmethionine 500 mg, trihydroxyethylrutose 500 mg, distilled water 15 ml, glycerol to 50g.

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62845R-M. M13. STEK. 07-03-69.
 NL-403615. R35.
 Kohn Philip. Macomber, NY. BE--74-017-Q...

C23c (31-07-70)...
 PASSIVATING STEEL SURFACE COATING

NEW

Method of treatment of the surface of a steel article by subjecting the steel article to the presence of a group of volatile element or a compound of a volatile element, so that the element is incorporated into the surface of the steel article.

SPECIFICALLY

An iron compound of the element is applied to the surface of the article and allowed to diffuse into the article by annealing. The iron compound may be applied by evaporating, condensing, spraying, by electrolytic method, or by coating with a slurry. The annealing takes place under vacuum or in an inert atmosphere for one hour. The steel article is then annealed in an atmosphere containing a volatile compound of the element, such as an oxide or an organic compound. The atmosphere prior consists of argon containing 10% of the volatile compound.

M13-H.

296

USES/ADVANTAGES

The method facilitates the production of steel articles, especially articles for use in sodium-coated reactors. The treatment prevents carbon transport in the treated steel article. The treated article has considerably improved surface mechanical properties, and corrosion resistance.

EXAMPLE

An iron-niobium compound was applied as a slurry onto the surface of a pump fan composed of an unstabilized steel. The coated article was then annealed in an inert atmosphere for 1 hr. at 900 °C.

62845R